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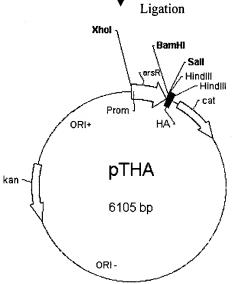
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7589 bp

Prom

Xhol

arsR

CTCGAG-----TGAGAAAAGGAGGCGGATCCATG---

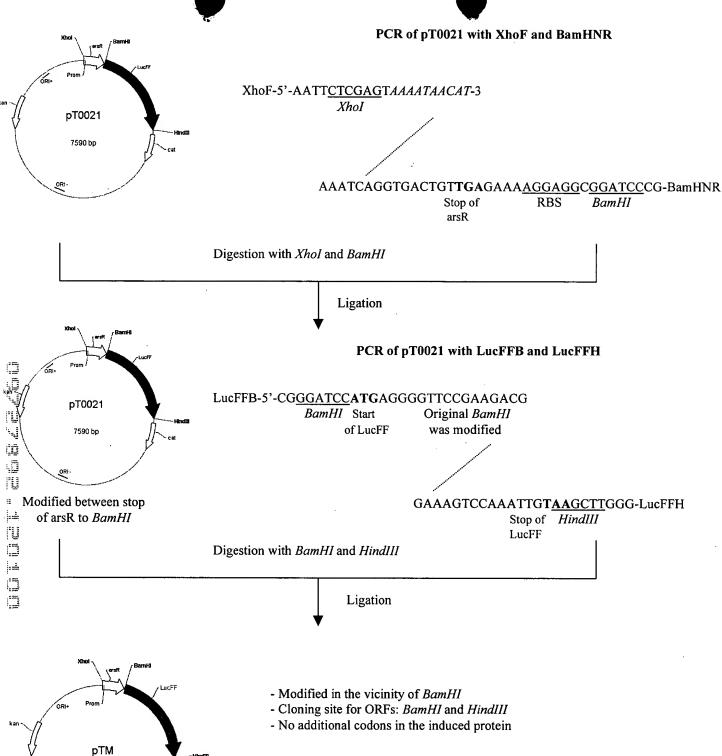
RBS

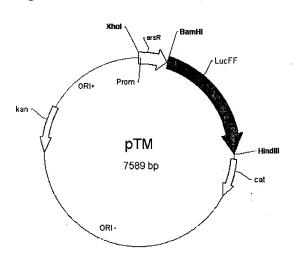
BamHI

TAAGCTT

HindIII

FIGURE 1B





CTCGAG--XhoI

PCR of Staphylococcus aureus DNA with lacF and LacR

LacF-5`-CCGCTCGAGCTCCAAATTCCAAAACAG
XhoI

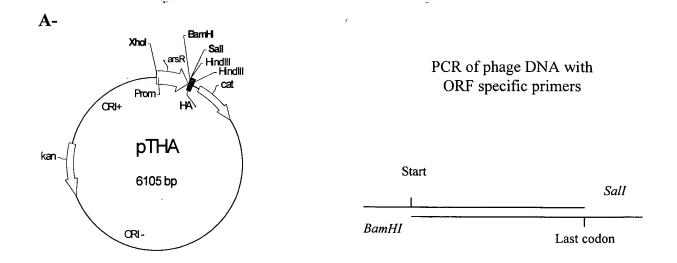
GTAAAAAGGAGTCTTATT<u>GGATCC</u>CG-LacR

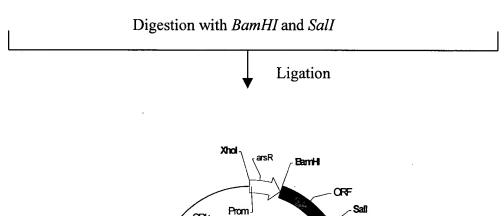
BamHI

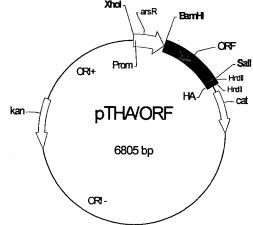
Digestion with Xhol and BamHI Ligation Xhol Prom Prom LacR Prom LacA Prom LacA

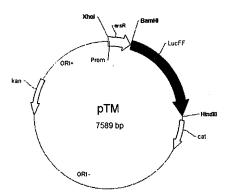
--TTG-----CTA-----AGGAGTCTTATTGGATCCATG

BamHI



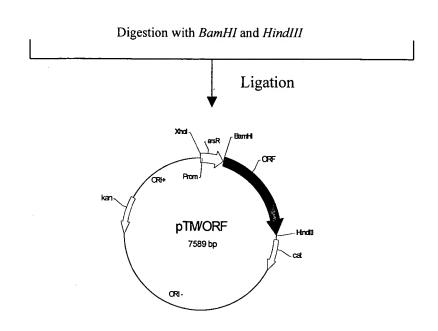






PCR of phage DNA with ORF specific primers





A) Functional assay on semi-solid support medium

O/N culture inoculated from frozen stock of phage ORF *Staphylococcus aureus* RN4220 transformants

1:10, 1:1000, 1:1000, 1:10000 dilution in saline solution

3 ul of dilution

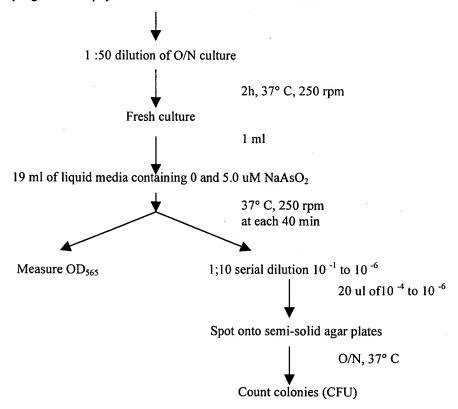
Spot on semi-solid support agar plates containing
0, 2.5, 5.0, 7.5 uM NaAsO₂

O/N, 37° C

Compare bacterial growth on plates with and without NaAsO₂

B) Functional assay in liquid medium

O/N culture inoculated from frozen stock of phage ORF *Staphylococcus aureus* RN4220 transformants

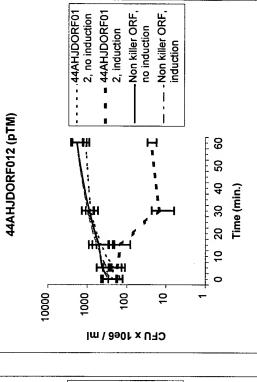


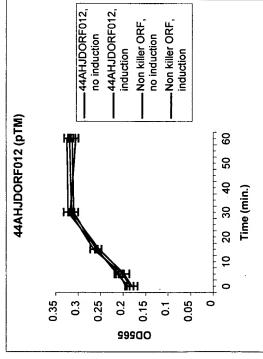
44AHJD ORFs tested for functional analysis

UID	POS
44AHJDORF001	1262710342
44AHJDORF002	37895732
44AHJDORF003	66268389
44AHJDORF004	876410227
44AHJDORF005	1389012643
44AHJDORF006	8032029
44AHJDORF007	20443027
44AHJDORF008	30203775
44AHJDORF009	57446496
44AHJDORF010	1442013938
44AHJDORF011	1559315225
44AHJDORF012	83918813
44AHJDORF013	1499614586
44AHJDORF019	98369630
44AHJDORF023	64946315
44AHJDORF025	1517514999
44AHJDORF027	1291613080
44AHJDORF028	92359071
44AHJDORF035	1395713811
44AHJDORF036	1016510019
44AHJDORF039	17431883
44AHJDORF040	97409877
44AHJDORF044	1291712783
44AHJDORF046	48915019
44AHJDORF048	1534015212
44AHJDORF049	57845909
44AHJDORF053	33483467
44AHJDORF113	199600
44AHJDORF114	1617215870
44AHJDORF121	1636216165
44AHJDORF123	614796

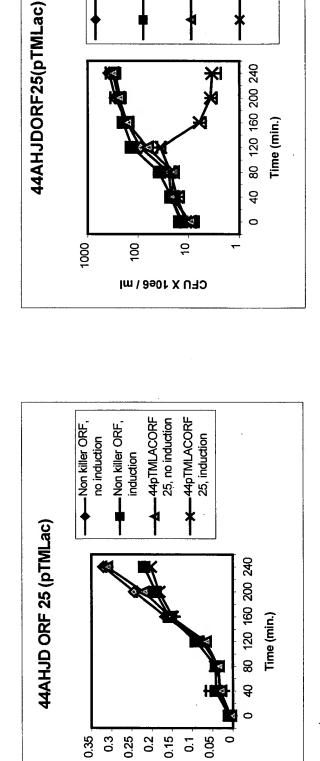
4	1

		support media
Staphylococcus aureus transformants	Without induction	With induction (5 uM sodium arsenite)
Clone1		
Clone2		
Clone3		
Clone1		
Clone2		
Clone3		
Clone1		0/0/4
	aureus transformants Clone1 Clone2 Clone3 Clone2 Clone3	transformants Clone1 Clone2 Clone3 Clone2 Clone3 Clone2 Clone3





Non killer ORF, no induction .44pTORF025, No induction Non killer ORF, induction -44pTORF025, Induction 44AHJDORF025 (pT) 240 500 120 Time (min.) 40 0.01 0.001 0.0001 CFU X 10e6 / ml Non killer, no 12 44pTORF25 No induction 44pTORF25 44AHJDORF025 (pT) ロコトロミト Non killer, induction induction induction 240 160 Time (min.) 8 0 **2 20** 4.0 0.3 0.4 0.2 0.1



OD265

-44pTMLAC ORF25, induction

Non killer ORF, no induction Non killer ORF, induction

-44pTMLAC ORF25, no induction

Fig.

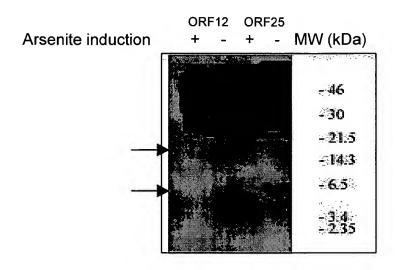
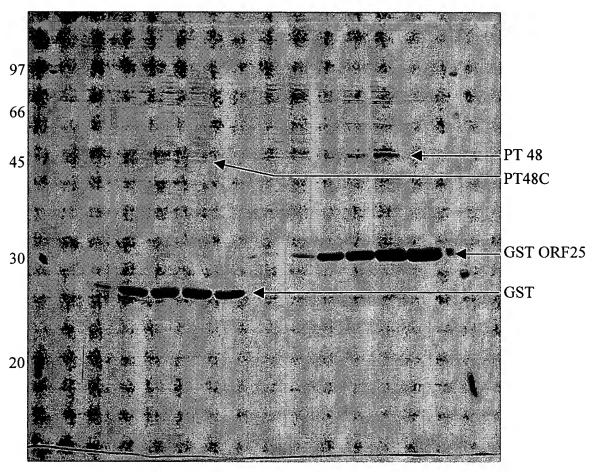


FIGURE 7A

GST ORF25
Mr 0 0.1 0.5 1.0 2.0 0 0.1 0.5 1.0 2.0 ACB

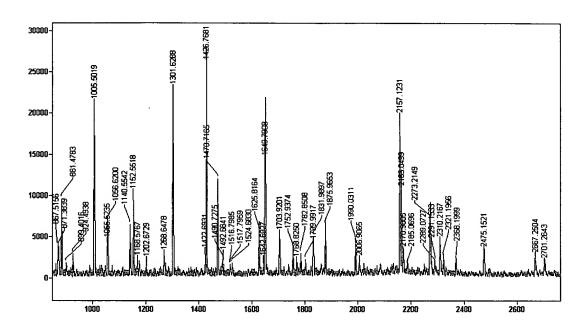
FIGURE 7B GST ORF25

Mr 0 0.1 0.5 1.0 2.0 ACB 0 0.1 0.5 1.0 2.0 ACB Mr



Tryptic peptide mass spectrum of 48 kDa interacting protein

(from Figure 1B)





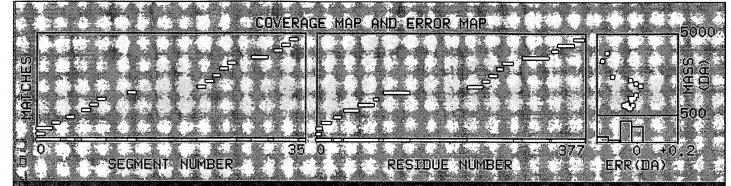


Identification of PT48 as DNA-directed DNA polymerase III beta.

Details for : PT48 gi|1706496|sp|P50029|DP3B_STAAU_DNA_POLYMERASE III, BETA CHAIN gi|1084187|pir||S54708 DNA-directed DNA polymerase (EC 2.7.7.7) III beta chain - Staphylococcus aureus

Sample ID : PT 48

Number of Measured Peptides : 40 Number of Matched Peptides : 17 Coverage of protein sequence : 68%



Measured	Avg/	Computed	Error	Resid	dues	Missed	
Mass(M)	Mono	Mass	(Da)	Start	То	Cut	Peptide sequence
	4.4	# 33 J	e • #	: A)		8.4	歌唱的 化甲磺基甲基磺基甲基甲基甲基甲基甲基甲基甲基甲基甲基甲基甲基甲基甲基甲基甲基甲基
s 870.370	M.	870.397	-0.027	333	339	0	YMMDALK
£880.474	The state of the s	200000 Teaching and the contract of the contra	St. P. Land Control of	Professional Committee of	88	0	FFVDIIK
#1898.392					7	0.	MMEFTIK
= 004.492					243	0	VGNVNFLSR
1054.514 1055.603					35	, O T	MMEFTIKR TTLPILTGIK
151.546	Mary Name of Street, or other	alling a stillet contillation, their	- A 2000 CAMBO A CAMBO	Broadshillian La Million and	200000 A. C. CONTROL		TEPENYEIK
4300.619	9.		2007	1 7	2004	2000 2000 1000	LLEGHYPDTTR
1425.769					375		GDDSVTQLTLPIR
1469.713					20		DYFITQLINDTLK
1648.781					277	200000 Yes	LSIDNGEFYHAIDR
2156.112 2309.198					90 81	- 0	TVDGEDIVNISETGSVVLPGR EHEVILEGSDSETSTETTIPK
2474.139				214			IMSDNEEDIDIFFASNOVLFK:
2666.266		-2005 COMPANY	- Administrative (2000)	340	ACRES TO A SECOND	CONTRACT OF THE PARTY OF THE PA	AIDNDEVEVEFFGIMKPFILKPK
3577.670	Ā	3577.809	-0.139	292			LSTGDDVVELSSTSPEIGTVKEEVDANDVEGGSLK
4033.298	Α	4033.421	-0.123	. 97	132	0	LSTNEOFOTLITSGHSEFNLSGLDPDQYPLLPQVSR



SEQ ID NO: 1 Staphyloccus aureus DnaN nucleotide sequence

ATGATGGAATTCACTATTAAAAGAGATTATTTTATTACACAATTAAATGACACATTAAAA GCTATTTCACCAAGAACAACATTACCTATATTAACTGGTATCAAAATCGATGCGAAAGAA CATGAAGTTATATTAACTGGTTCAGACTCTGAAATTTCAATAGAAATCACTATTCCTAAA ACTGTAGATGGCGAAGATATTGTCAATATTTCAGAAACAGGCTCAGTAGTACTTCCTGGA CGATTCTTTGTTGATATTATAAAAAAATTACCTGGTAAAGATGTTAAATTATCTACAAAT GAACAATTCCAGACATTAATTACATCAGGTCATTCTGAATTTAATTTAAGTGGCTTAGAT CCAGATCAATATCCTTTATTACCTCAAGTTTCTAGAGATGACGCAATTCAATTGTCGGTA AAAGTGCTTAAAAACGTGATTGCACAAACAACTTTTGCAGTGTCCACCTCAGAAACACGC CCAGTACTAACTGGTGTGAACTGGCTTATACAAGAAAATGAATTAATATGCACAGCGACT GACTCACACCGCTTGGCTGTAAGAAAGTTGCAGTTAGAAGATGTTTCTGAAAACAAAAAT GTCATCATTCCAGGTAAGGCTTTAGCTGAATTAAATAAAATTATGTCTGACAATGAAGAA ATTTCTCGATTATTAGAAGGACATTATCCTGATACAACACGTTTATTCCCTGAAAACTAT GAAATTAAATTAAGTATAGACAATGGGGAGTTTTATCATGCGATTGATCGTGCCTCTTTA TTAGCGCGTGAAGGTGGTAATAACGTTATTAAATTAAGTACAGGTGATGACGTTGTTGAA TTGTCTTCTACATCACCAGAAATTGGTACTGTAAAAGAAGAAGTTGATGCAAACGATGTT ATCGATAATGATGAGGTTGAAGTTGAATTCTTCGGTACAATGAAACCATTTATTCTAAAA CCAAAAGGTGACGACTCGGTAACGCAATTAATTTTACCAATCAGAACTTACTAA

SEQ ID NO: 2 Staphyloccus aureus DnaN amino acid sequence

>gi|1084187|pir||S54708 DNA-directed DNA polymerase (EC 2.7.7.7) III beta chain - Staphylococcus aureus

MMEFTIKRDYFITQLNDTLKAISPRTTLPILTGIKIDAKEHEVILTGSDSEISIEITIPKTVDGEDIVNI SETGSVVLPGRFFVDIIKKLPGKDVKLSTNEQFQTLITSGHSEFNLSGLDPDQYPLLPQVSRDDAIQLSV KVLKNVIAQTNFAVSTSETRPVLTGVNWLIQENELICTATDSHRLAVRKLQLEDVSENKNVIIPGKALAE LNKIMSDNEEDIDIFFASNQVLFKVGNVNFISRLLEGHYPDTTRLFPENYEIKLSIDNGEFYHAIDRASL LAREGGNNVIKLSTGDDVVELSSTSPEIGTVKEEVDANDVEGGSLKISFNSKYMMDALKAIDNDEVEVEF FGTMKPFILKPKGDDSVTQLILPIRTY